pB1

287 Bfr BI
287 Ns I
287 Ppu 10I

Sca I 6720

Bpm I 6327

1285 Xcm I
1288 Bse RI

AlwNI 5760

pB1
+
7159 base pairs
Unique Sites

Sap I 6226

1748 Bsp EI
1841 Pml I
1956 Apa BI
1956 Bsl API
2124 Bsu 36I
2174 Bst XI

Pst I 4776
Sal I 4770
BamHI 4764
Xma I 4759
Sma I 4759
Sty I 4749
Nco I 4749
Bst DSI 4749
Sfi I 4747
Msc I 4746
Hpa I 4570

Bst BI 4026
Bsa BI 3797

2880 Dra III
2986 Nae I
2986 Ngo MIV

3289 Nru I
3293 Ecl 136II
3293 Sac I
3384 Mlu I
3465 Ppu MI
3515 Rle AI

Oligo 160

| gagagtagtaacaaaggtc | AAAGACAGTTGACTGTATCGCCG GAA TTT AT

           Sfi I   Sma I   BamH I   Sal I   Pst I

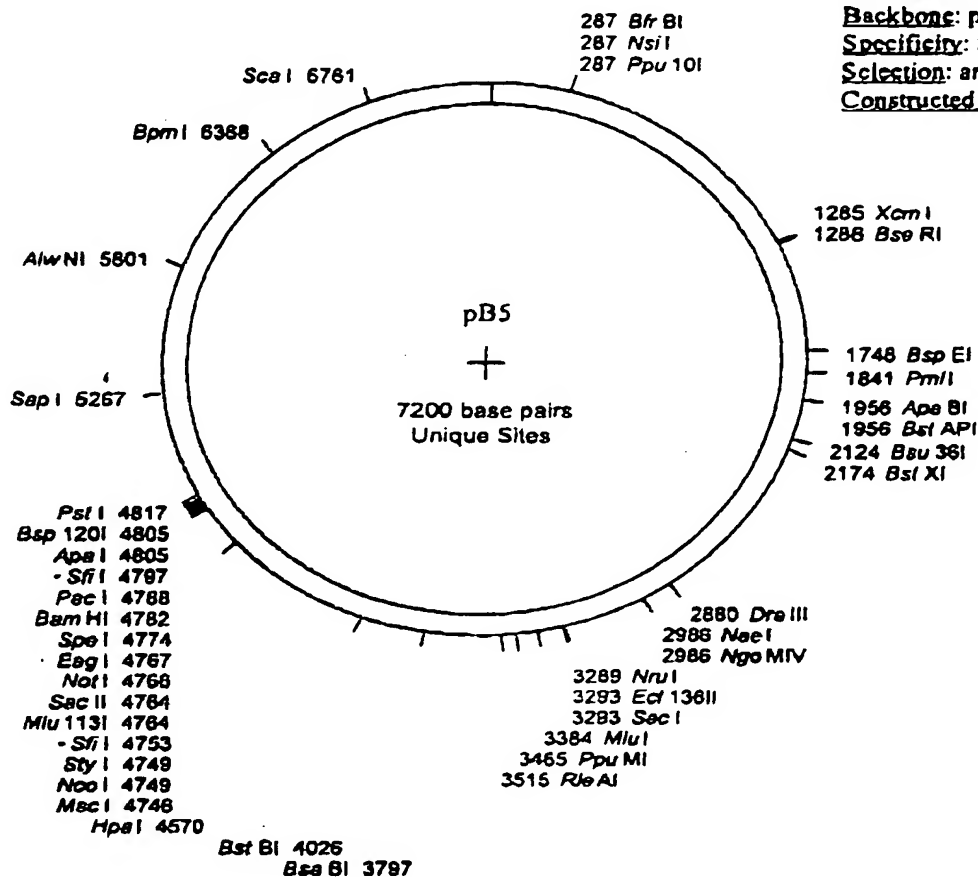G GCC ATG GAG GCC CCG GGG ATC CGT CGA CCT GCA GCC

        Nco I

Oligo 161

AAG CTA ATT | ccgggcgaattcttatg |

Oligo 160 5' GAGAGTAGTAACAAAGGTC 3'
Oligo 161 5' CATAAGAAATTCGCCCGG 3'

FIGURE 1

pB5[2]

287 Bfr BI
287 Nsi I
287 Ppu 10I

Sca I 6781

Bpm I 6388

1285 Xcm I
1288 Bse RI

AlwNI 5801

pB5

+

7200 base pairs
Unique Sites

Sap I 5267

1748 Bsp EI
1841 Pml I
1956 Ape BI
1956 Bsi API
2124 Bsu 36I
2174 Bst XI

Pst I 4817
Bsp 120I 4805
Apa I 4805
- Sfi I 4797
Pac I 4788
Bam HI 4782
Spe I 4774
Eag I 4767
Not I 4768
Sac II 4764
Mlu 113I 4764
- Sfi I 4753
Sty I 4749
Nco I 4749
Msc I 4748
Hpa I 4570

2880 Dra III
2986 Nae I
2986 Ngo MIV

3289 Nru I
3293 Ecl 136II
3283 Sac I
3384 Mlu I
3465 Ppu MI
3515 Rle AI

Bst BI 4026
Bsa BI 3797

Oligo 160

| gagagtagtaacaaaggtc | AAAGACAGTTGACTGTATCGCCG GAA TTT ATG

| | | Sfi I | | Sac II | | | Spe I | | Bam HI |
GCC ATG GCC GCA GGG GCC GCG GCC GCA CTA GTG GGG ATC C
Nco I | | | | | Not 1

| | STOP | | Sfi I | | | Pst I |
TT AAT TAA GGG CCA CTG GGG CCC CTC GAC CTG CAG CCA
Pac I

Oligo 161
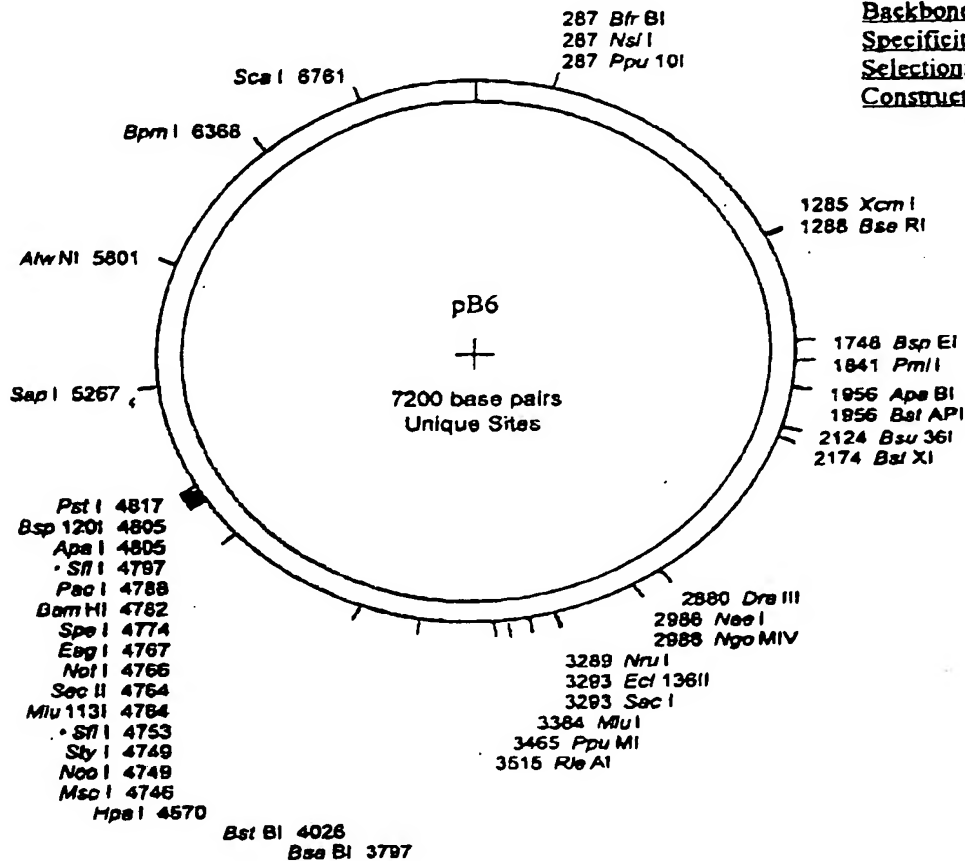
AGC TAA TT ccgggcgaatttcttatg

Oligo 160 5' GAGAGTAGTAACAAAGGTC 3'
Oligo 161 5' CATAAGAAATTCGCCCGG 3'

FIGURE 2

pB6 [3]

```
                                    287 Bfr BI
                                    287 Nsi I
                                    287 Ppu 10I
        Sca I 6761

    Bpm I 6368
                                                    1285 Xcm I
                                                    1288 Bse RI
    Afw NI 5801

                            pB6                     1748 Bsp EI
                             +                      1841 Pml I
                        7200 base pairs             1956 Apa BI
    Sap I 5267          Unique Sites                1956 Bst API
                                                    2124 Bsu 36I
                                                    2174 Bst XI
    Pst I 4817
    Bsp 1201 4805
    Apa I 4805
    · Sfi I 4797                                2880 Dra III
    Pac I 4788                                  2986 Nae I
    Bam HI 4782                                 2988 Ngo MIV
    Spe I 4774                          3289 Nru I
    Eag I 4767                          3293 Ecl 136II
    Not I 4766                          3293 Sac I
    Sec II 4764                         3384 Mlu I
    Mlu 113I 4764                       3465 Ppu MI
    · Sfi I 4753                        3515 Rle AI
    Sty I 4749
    Nco I 4749
    Msc I 4746
    Hpa I 4570
            Bst BI 4026
              Bse BI 3797
```

Oligo 160

| gagagtagtaacaaaggtc |AAAGACAGTTGACTGTATCGCCG GAA TTT ATG

	Sfi I	Sac II		Spe I		Bam HI

GCC ATG GCC GGA CGG GCC GCG GCC GCA CTA GTG GGG ATC C

Nco I   Not I

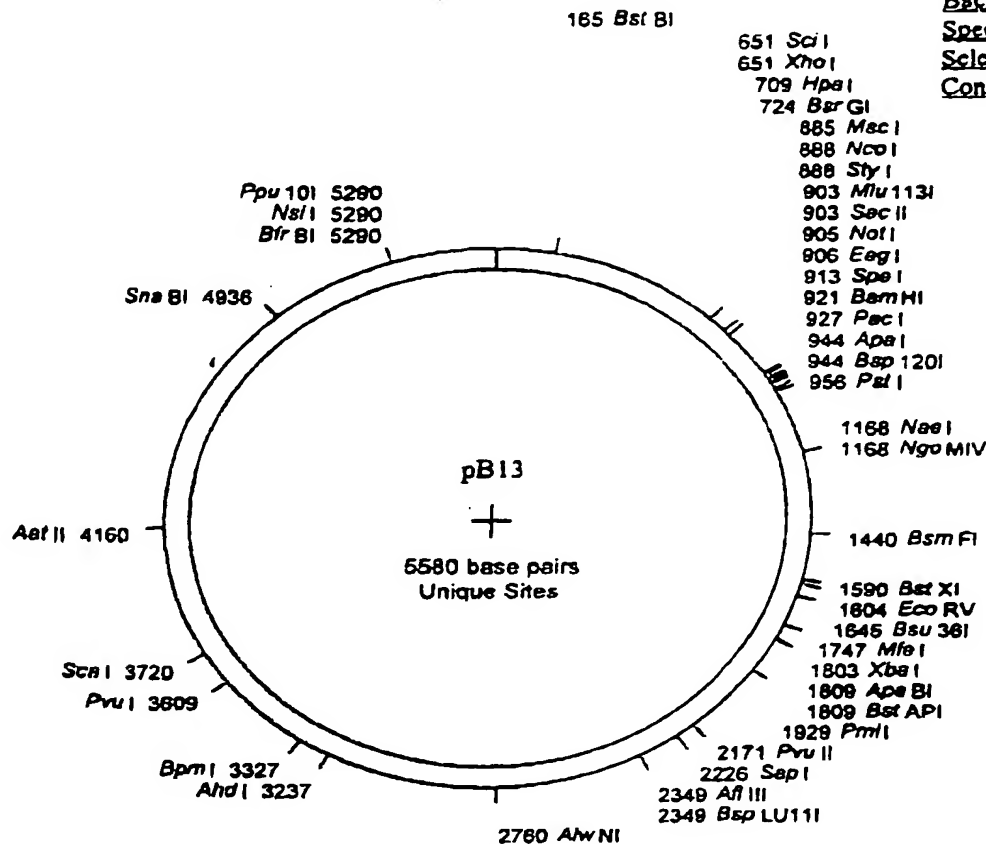|	STOP		Sfi I	Apa I		Pst I

TT AAT TAA GGG CCA CTG GGG CCC CTC GAC CTG CAG CCA

Pac I

Oligo 161

AGC TAA TT ccgggcgaatttcttatg

Oligo 160 5' GAGAGTAGTAACAAAGGTC 3'
Oligo 161 5' CATAAGAAATTCGCCCGG 3'

FIGURE 3

pB13

165 *Bst* BI

651 *Sci* I
651 *Xho* I
709 *Hpa* I
724 *Bsr* GI
885 *Msc* I
888 *Nco* I
888 *Sty* I
903 *Mlu* 113I
903 *Sac* II
905 *Not* I
906 *Eag* I
913 *Spe* I
921 *Bam* HI
927 *Pac* I
944 *Apa* I
944 *Bsp* 120I
956 *Pst* I

Ppu 10I 5290
Nsi I 5290
Bfr BI 5290

Sna BI 4936

1168 *Nae* I
1168 *Ngo* MIV

Aat II 4160

pB13
+
5580 base pairs
Unique Sites

1440 *Bsm* FI

1590 *Bst* XI
1604 *Eco* RV
1645 *Bsu* 36I
1747 *Mfe* I
1803 *Xba* I
1809 *Ape* BI
1809 *Bst* API
1929 *Pml* I

Sca I 3720
Pvu I 3609

2171 *Pvu* II
2226 *Sap* I
2349 *Afl* III
2349 *Bsp* LU11I

Bpm I 3327
Ahd I 3237

2760 *Alw* NI

Oligo 160

gagagtagtaacaaaggtc AAAGACAGTTGACTGTATCGCCG GAA TTT ATG

| | Sfi I | | | Sac II | | | Spe I | | | Bam HI | |
GCC ATG GCC GCA GGG GCC GCG GCC GCA CTA GTG GGG ATC C
Nco I   Not I

STOP   Sfi I   Pst I
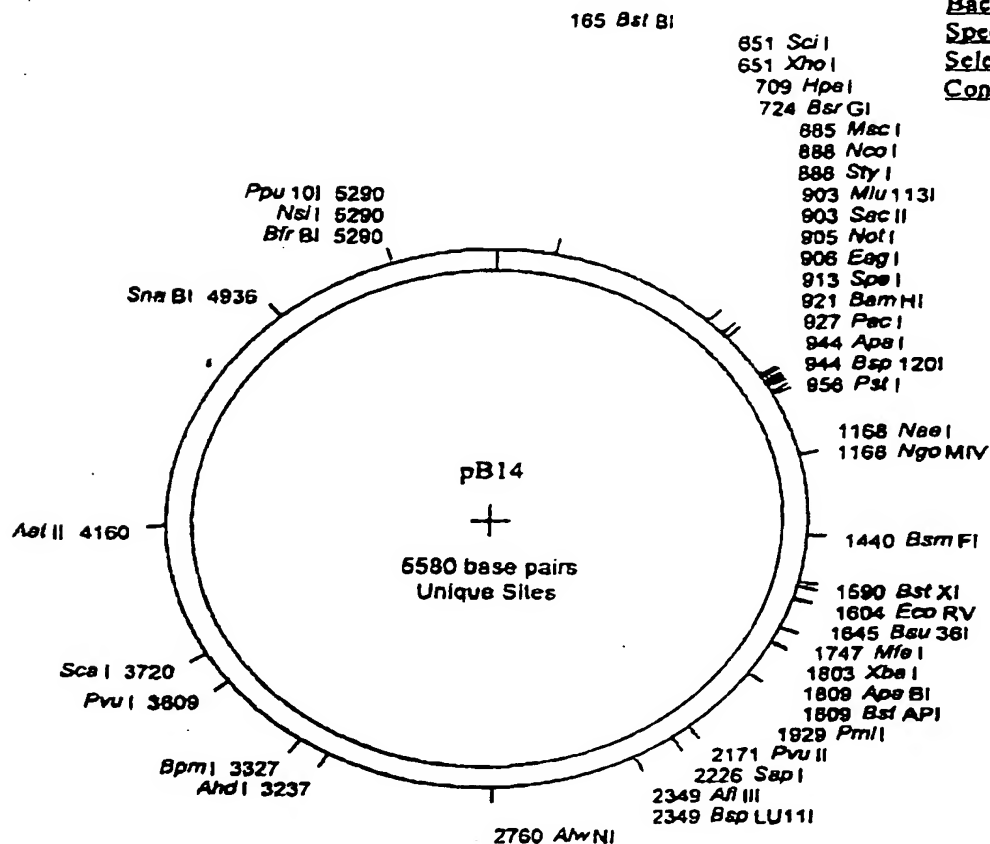TT AAT TAA GGG CCA CTG GGG CCC CTC GAC CTG CAG CCA
Pac I

Oligo 161
AGC TAA TT ccgggcgaatttcttatg

Oligo 160 5' GAGAGTAGTAACAAAGGTC 3'
Oligo 161 5' CATAAGAAATTCGCCCGG 3'

FIGURE 4

pB14

165 *Bst* BI

651 *Sci* I
651 *Xho* I
709 *Hpa* I
724 *Bsr* GI
885 *Msc* I
888 *Nco* I
888 *Sty* I
903 *Mlu* 113I
903 *Sac* II
905 *Not* I
906 *Eag* I
913 *Spe* I
921 *Bam* HI
927 *Pac* I
944 *Apa* I
944 *Bsp* 120I
958 *Pst* I

1168 *Nae* I
1168 *Ngo* MIV

1440 *Bsm* FI

1590 *Bst* XI
1604 *Eco* RV
1645 *Bsu* 36I
1747 *Mfe* I
1803 *Xba* I
1809 *Ape* BI
1809 *Bst* API
1929 *Pml* I
2171 *Pvu* II
2226 *Sap* I
2349 *Afl* III
2349 *Bsp* LU11I

Ppu 10I 5290
Nsi I 5290
Bfr BI 5290

Sna BI 4936

Aat II 4160

pB14
+
5580 base pairs
Unique Sites

Sca I 3720
Pvu I 3809

Bpm I 3327
Ahd I 3237

2760 *Alw* NI

Oligo 160

| gagagtagtaacaaaggtc | AAAGACAGTTGACTGTATCGCCG GAA TTT ATG

           Sfi I   Sac II   Spe I   Bam HI

GCC ATG GCC GGA CGG GCC GCG GCC GCA CTA GTG GGG ATC C
   Nco I   Not I

     STOP   Sfi I Apa I   Pst I

TT AAT TAA GGG CCA CTG GGG CCC CTC GAC CTG CAG CCA
   Pac I

Oligo 161

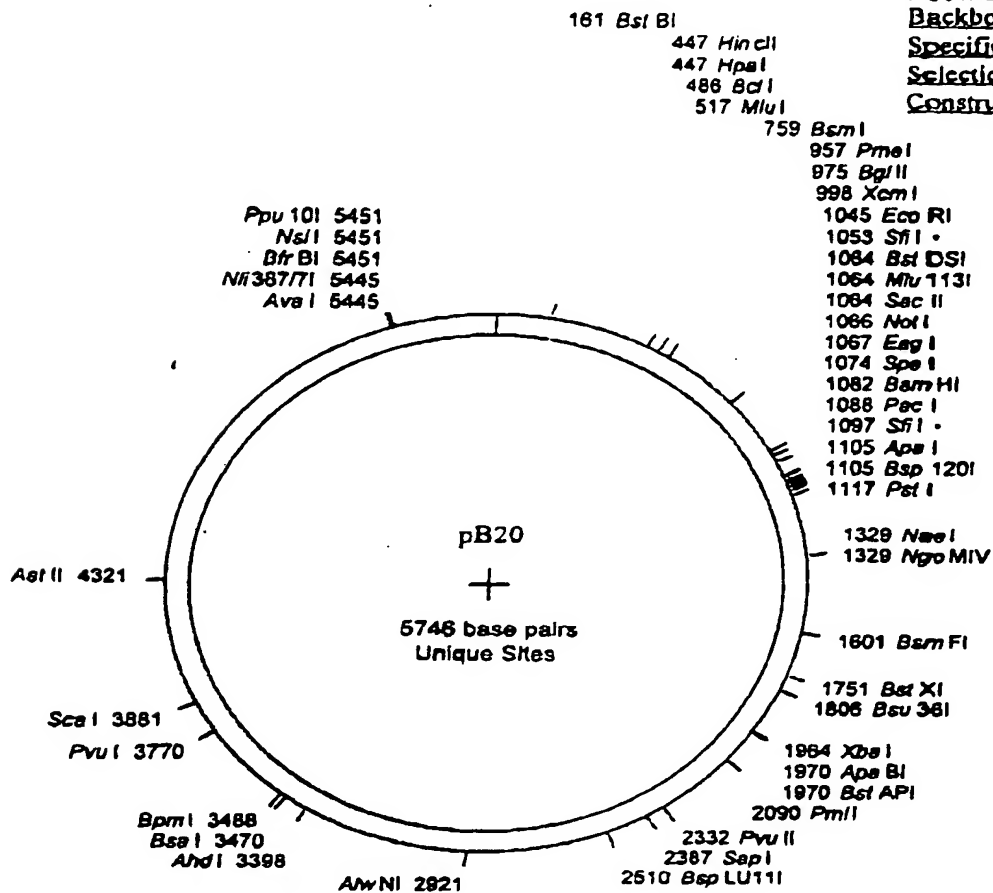AGC TAA TT ccgggcgaatttcttatg

Oligo 160 5' GAGAGTAGTAACAAAGGTC 3'
Oligo 161 5' CATAAGAAATTCGCCCGG 3'

FIGURE 5

pB20

Alias: pLex10NS2
Application: 2HY (bait)
Backbone: pLex10 (pB9)
Specificity: Sfi-oriented
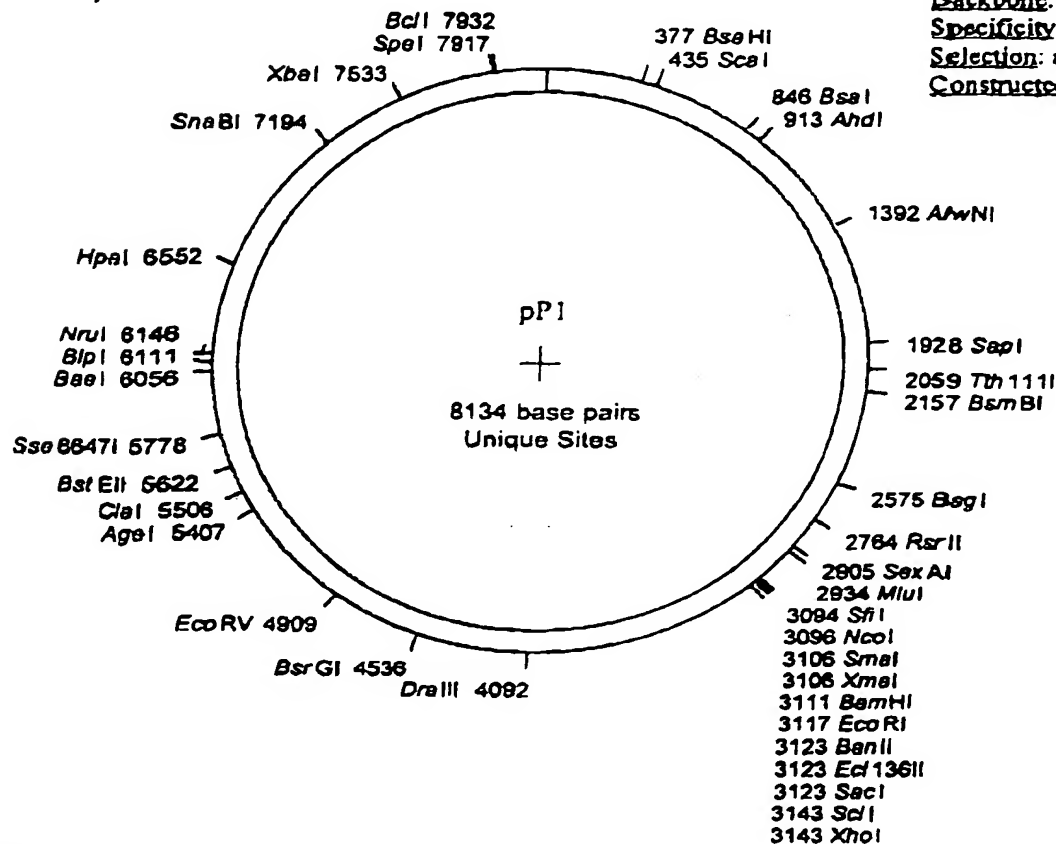Selection: ampicillin
Constructed by: LD

161 *Bst* BI
447 *Hin* cII
447 *Hpa* I
486 *Bcl* I
517 *Mlu* I
759 *Bsm* I
957 *Pme* I
975 *Bgl* II
998 *Xcm* I
1045 *Eco* RI
1053 *Sfi* I ·
1064 *Bst* DSI
1064 *Mlu* 113I
1064 *Sac* II
1086 *Not* I
1067 *Eag* I
1074 *Spe* I
1082 *Bam* HI
1088 *Pac* I
1097 *Sfi* I ·
1105 *Apa* I
1105 *Bsp* 120I
1117 *Pst* I

1329 *Nae* I
1329 *Ngo* MIV

1601 *Bsm* FI

1751 *Bst* XI
1806 *Bsu* 36I

1984 *Xba* I
1970 *Apa* BI
1970 *Bst* API
2090 *Pml* I

2332 *Pvu* II
2387 *Sap* I
2510 *Bsp* LU11I

Ahw NI 2921

Bpm I 3488
Bsa I 3470
Ahd I 3398

Pvu I 3770

Sca I 3881

Aat II 4321

Ppu 10I 5451
Nsi I 5451
Bfr BI 5451
Nli 387/TI 5445
Ava I 5445

pB20

+

5746 base pairs
Unique Sites

EcoR I		Sfi I				Not I		Spe I		BamH I	
GAA	TTC	GGG	GCC	GGA	CGG	GCC	GCG	GCC	GCA	CTA	GTG	GGG	ATC	C

Sac II

STOP

			Sfi I						Pst I	
TT	AAT	TAA	GGG	CCA	CTG	GGG	CCC	CTC	GAC	CTG	CAG

Pac I

FIGURE 6

pP1

Bcll 7932
Spel 7917
Xbal 7533
SnaBI 7194

377 Bsa HI
435 Scal
846 Bsa I
913 Ahdl

1392 AlwNI

Hpal 6552

Nrul 6146
Blpl 6111
Bael 6056

pP1

8134 base pairs
Unique Sites

1928 Sapl
2059 Tth 111I
2157 BsmBI

Sse 8647I 5778
Bst EII 5622
Clal 5506
Agel 5407

2575 Bsgl
2764 RsrII
2905 SexAI
2934 Mlul
3094 Sfil
3096 Ncol
3106 Smal
3106 Xmal
3111 BamHI
3117 EcoRI
3123 BanII
3123 Ecl136II
3123 Sacl
3143 Scil
3143 Xhol

EcoRV 4909
BsrGI 4536
DraIII 4092

ABS1

| cgtttggaatcactacagg | GATGTTTAATACCACTACAATGGATGATGTATATAACTATCTATT |

JC90

                          Bgl II

| cgatgatgaagatacccaccaaa | CCCAAAAAAAGAGATCTGTATGGCTTACCCATACGATGTTCCAG |

                          Sfi I   **Sma I**   **BamH I**

ATTACGCTAGCTTGGGTGGTCATATGGCC ATG GAG GCC CCG GGG ATC CGA ATT

                               Nco I
Sac I   **Xho I**   **Bgl II**

CGA GCT CGA CTA GCT AGC TGA CTC GAG AGA TCT ATGAAT

| cgtagatactgaaaaaccoc | GCAAGTT | cacttcaactgtgcatcgtg | caccatctcaatttc |

    162   **ABS2**   **53**

ABS1 5' CGTTTGGAATCACTACAGG 3'
JC90 5' CGATGATGAAGATACCCACCAAA 3'
162   5' GGGGTTTTTCAGTATCTACG 3'
ABS2 5' CACGATGCACAGTTGAAGTG 3'
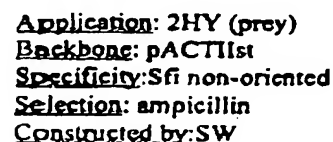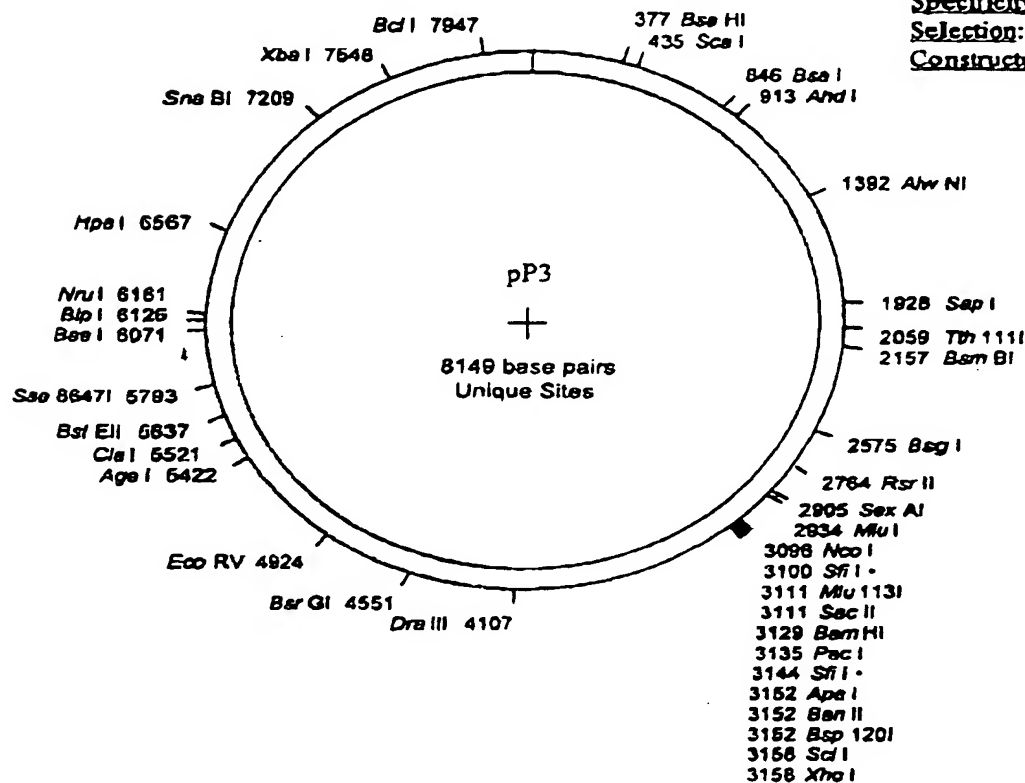53   5' GAAATTGAGATGGTGCACGATGCAC 3'

FIGURE 7

pP2[8]

Bcl I 7947
Xba I 7548
Sna BI 7209
Hpa I 6567
Nru I 6161
Blp I 6126
Bae I 6071
Sse 8647I 5793
Bst EII 5637
Cla I 5521
Age I 5422
Eco RV 4924
Bsr GI 4651
Dra III 4107

377 Bsa HI
435 Sca I
846 Bsa I
913 Ahd I
1392 Ahr NI
1928 Sap I
2059 Tth 111I
2157 Bsm BI
2575 Bsg I
2764 Rsr II
2905 Ser AI
2934 Mlu I
3096 Nco I
3100 Sfi I ·
3111 Mlu 113I
3111 Sac II
3129 Bam HI
3135 Pac I
3144 Sfi I ·
3152 Apa I
3152 Ban II
3152 Bsp 120I
3158 Sci I
3158 Xho I

pP2
+
8149 base pairs
Unique Sites

ABS1

CG [cgtttggaatcactacagg] GATGTTTAATACCACTACAATGGATGATGTATATAACTATCTATT

JC90

[cgatgatgaagatacccaccaaa] CCCAAAAAAAGAGATCTGTATGGCTTACCCATACGATGTTCCAG

Bgl II

Sfi I   Sac II

ATTACGCTAGCTTGGGTGGTCATATGGCC ATG GCC GCA GGG GCC GCG GCC GCA.
Nco I

BamH I   Pac I

CTA GTG GGG ATC CTT AAT [TAA] GGG CCA CTG GGG CCC CTC GAG AGA TCT
Stop

ATGAAT [cgtagatactgaaaaacccc] GCAAGTT [cacttcaact][gtgcatcgtg][caccatctcaatttc]

162   ABS2   53

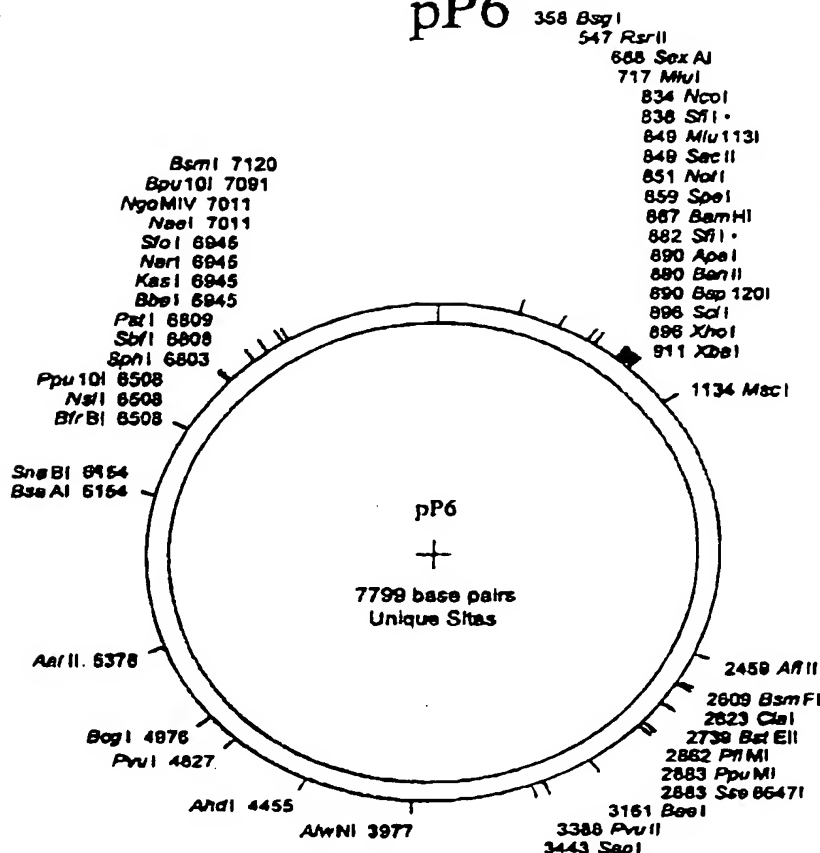ABS1 5' CGTTTGGAATCACTACAGG 3'
JC90 5' CGATGATGAAGATACCCCACCAAA 3'
162   5' GGGGTTTTTCAGTATCTACG 3'
ABS2 5' CACGATGCACAGTTGAAGTG 3'
53   5' GAAATTGAGATGGTGCACGATGCAC 3'

FIGURE 8

pP3[9]

Bcl I 7947
Xba I 7648
Sna BI 7209
Hpa I 6567
Nru I 6161
Blp I 6126
Bsa I 6071
Sse 86471 5793
Bst EII 5637
Cla I 5521
Age I 5422
Eco RV 4924
Bsr GI 4551
Dra III 4107

377 Bss HI
435 Sca I
846 Bsa I
913 Ahd I
1392 Alw NI
1928 Sap I
2059 Tth 111I
2157 Bsm BI
2575 Bsg I
2764 Rsr II
2905 Sex AI
2934 Mlu I
3096 Nco I
3100 Sfi I ·
3111 Mlu 113I
3111 Sac II
3129 Bam HI
3135 Pac I
3144 Sfi I ·
3152 Apa I
3152 Ban II
3152 Bsp 120I
3156 Scl I
3158 Xho I

pP3
+
8149 base pairs
Unique Sites

ABS1

CG [cgtttggaatcactacagg] GATGTTTAATACCACTACAATGGATGATGTATATAACTATCTATT

JC90   Bgl II

[cgatgatgaagataccccaccaaa] CCCAAAAAAAGAGATCTGTATGGCTTACCCATACGATGTTCCAG

                                    Sfi I   Sac II

ATTACGCTAGCTTGGGTGGTCATATGGCC ATG GCC GGA CGG GCC GCG GCC GCA
                              Nco I

        BamH I   Pac I

CTA GTG GGG ATC CTT AAT [TAA] GGG CCA CTG GGG CCC CTC GAG AGA TCT
                         Stop

ATGAAT [cgtagatactgaaaaacccc] GCAAGTT [cacttcaact][gtgcatcgtg] [caccatctcaatttc]

        162   ABS2   53

ABS1 5' CGTTTGGAATCACTACAGG 3'
JC90 5' CGATGATGAAGATACCCCACCAAA 3'
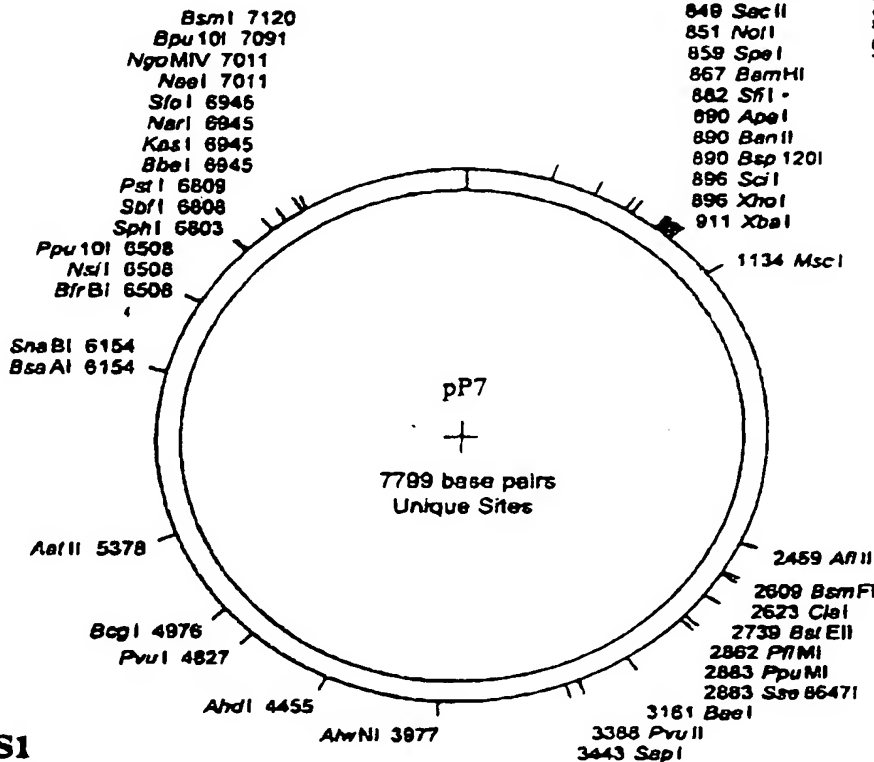162   5' GGGGTTTTTCAGTATCTACG 3'
ABS2 5' CACGATGCACAGTTGAAGTG 3'
53   5' GAAATTGAGATGGTGCACGATGCAC 3'

FIGURE 9

pP6

358 Bsg I
547 Rsr II
688 Sex AI
717 Mlu I
834 Nco I
838 Sfi I •
849 Mlu 113I
849 Sac II
851 Not I
859 Spe I
867 Bam HI
882 Sfi I •
890 Apa I
890 Ban II
890 Bsp 120I
896 Sal I
896 Xho I
911 Xba I

1134 Msc I

Alias: pGAD3S2XNS1
Application: 2HY (prey)
Backbone: pGAD3S2X
Specificity: Sfi non-oriented
Selection: ampicillin
Constructed by: SW

Bsm I 7120
Bpu10I 7091
NgoMIV 7011
Nae I 7011
Sfo I 6945
Nar I 6945
Kas I 6945
Bbe I 6945
Pst I 6809
Sbf I 6809
Sph I 6803
Ppu10I 6508
Nsi I 6508
Bfr BI 6508

Sna BI 6154
Bsa AI 6154

pP6

7799 base pairs
Unique Sites

Aat II. 5378

Bcg I 4876
Pvu I 4827

Ahd I 4455

AlwN I 3977

2459 Afl II
2609 Bsm FI
2623 Cla I
2739 Bst EII
2882 Pfl MI
2883 Ppu MI
2883 Sse 8647I
3161 Bce I
3388 Pvu II
3443 Sap I

ABS1

| cgtttggaatcactacagg | GATGTTTAATACCACTACAATGGATGATGTATATAACTATCTATT |

JC90

| cgatgatgaagatacccaccaaa | CCCAAAAAAAGAGATCCTAGAACTA |

Sfi I — Sac II — Spe I — Bam HI

GCC ATG GCC GCA GGG GCC GCG GCC GCA CTA GTG GGG ATC C
Nco I — Not I

STOP — Sfi I — Xho I — Xba I

TT AAT [TAA] GGG CCA CTG GGG CCC CTC GAG [TAG] C[TA G]TG T[CT AG]A
        STOP   STOP STOP   STOP

GGCCCGGTACCCAATTCGCCCTATAGTGAGTCGTATTACAATTCACTGGCCG TCGTTTTA

CAACGTCGTGACTGGGAAAACCCTGATCTATGAAT | cgtagatactgaaaaacccc | GCAA

162

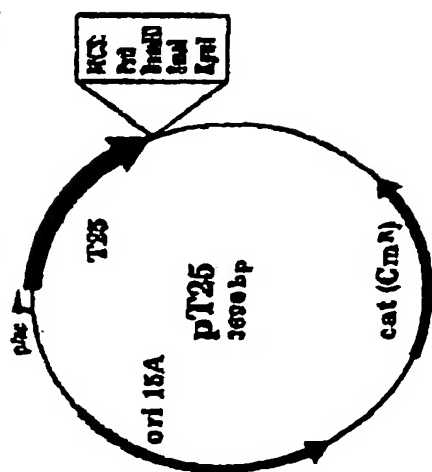GTT | cacttcaactgtgcatcgtg | caccatctcaatttctttc |
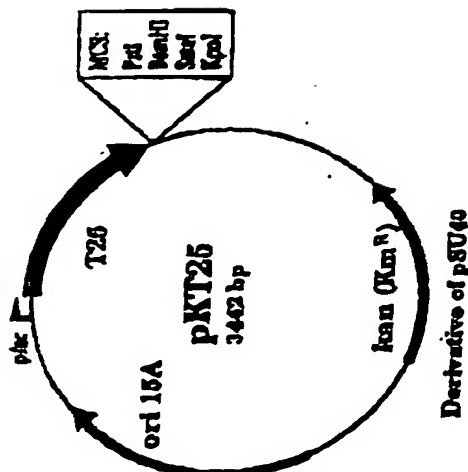
ABS2 — 53

ABS1 5' CGTTTGGAATCACTACAGG 3'
JC90 5' CGATGATGAAGATACCCCACCAAA 3'
162   5' GGGGTTTTTCAGTATCTACG 3'
ABS2 5' CACGATGCACAGTTGAAGTG 3'
53   5' GAAATTGAGATGGTGCACGATGCAC 3'

FIGURE 10

pP7

358 *Bsg* I
547 *Rsr* II
688 *Sex* AI
717 *Mlu* I
834 *Nco* I
838 *Sfi* I ·
849 *Mlu* 113I
849 *Sac* II
851 *Not* I
859 *Spe* I
867 *Bam* HI
882 *Sfi* I ·
890 *Apa* I
890 *Ban* II
890 *Bsp* 120I
896 *Sci* I
896 *Xho* I
911 *Xba* I

1134 *Msc* I

Alias: pGAD3S2XNS2
Application: 2HY (prey)
Backbone: pGAD3S2X
Specificity: Sfi oriented
Selection: ampicillin
Constructed by: SW

Bsm I 7120
Bpu 10I 7091
Ngo MIV 7011
Nae I 7011
Sfo I 6946
Nar I 6945
Kas I 6945
Bbe I 6945
Pst I 6809
Sbf I 6808
Sph I 6803
Ppu 10I 6508
Nsi I 6508
Bfr BI 6508

Sna BI 6154
Bsa AI 6154

Aat II 5378

Bcg I 4976
Pvu I 4827

Ahd I 4455

Ahw NI 3977

pP7

7799 base pairs
Unique Sites

2459 *Afl* II
2609 *Bsm* FI
2623 *Cla* I
2739 *Bst* EII
2862 *Pfl* MI
2883 *Ppu* MI
2883 *Sse* 8647I
3161 *Bae* I
3388 *Pvu* II
3443 *Sap* I

ABS1

| cgtttggaatcactacagg | GATGTTTAATACCACTACAATGGATGATGTATATAACTATCTATT

JC90

| cgatgatgaagatacoccaccaaa | CCCAAAAAAAGAGATCCTAGAACTA

            __Sfi I__   __Sac II__   __Spe I__   __Bam HI__

GCC ATG GCC GGA CGG GCC GCG GCC GCA CTA GTG GGG ATC C
      Nco I   **Not I**

    STOP   __Sfi I__   __Xho I__   **Xba I**

TT AAT TAA GGG CCA CTG GGG CCC CTC GAG TAG CTA GTG TCT AGA
                                    STOP   **STOP**   **STOP**

GGCCCGGTACCCAATTCGCCCTATAGTGAGTCGTATTACAATTCACTGGCCGTCGTTTTA

CAACGTCGTGACTGGGAAAACCCTGATCTATGAAT | cgtagatactgaaaaacccc | GCAA
                                                          162

GTT | cacttcaac | gtgcatcgtg | caccatctcaatttcttt |
           ABS2   **53**

ABS1 5' CGTTTGGAATCACTACAGG 3'
JC90 5' CGATGATGAAGATACCCCACCAAA 3'
162   5' GGGGTTTTTCAGTATCTACG 3'
ABS2 5' CACGATGCACAGTTGAAGTG 3'
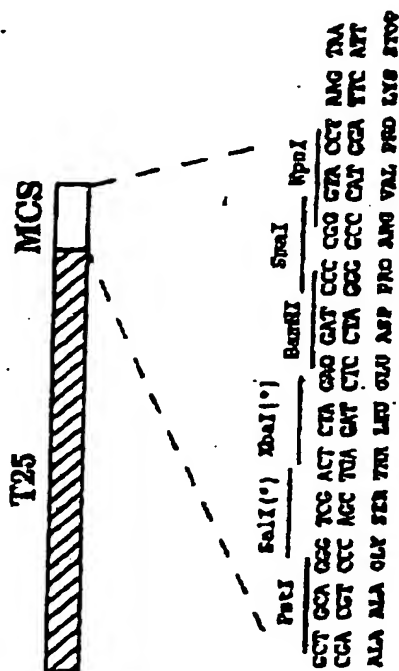53   5' GAAATTGAGATGGTGCACGATGCAC 3'
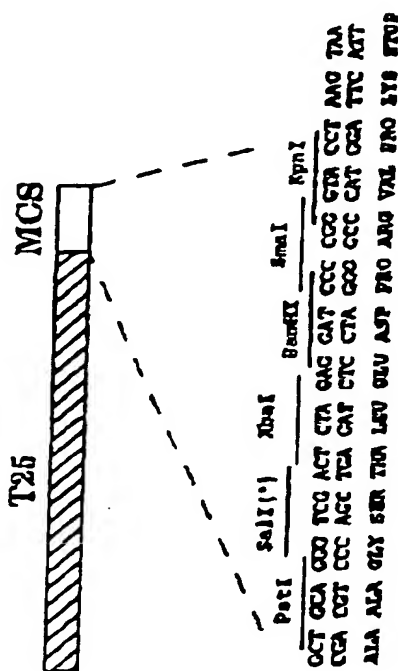
FIGURE 11

FIGURE 12

VECTORS EXPRESSING THE T25 FRAGMENT

FIGURE 12

VECTORS EXPRESSING THE T18 FRAGMENT

MCS   T18

Lac Z'

```
ATG ACC ATG ATT ACG CCA AGC TTG CAT GCC TCG ACT CTA GAG GAT CCC CGG GTA CCG AGC TCG AAT TCA
TAC TGG TAC TAA TGC GGT TCG AAC GTA CGG AGC TGA GAT CTC CTA GGG GCC CAT GGC TCG AGC TTA AGT
MET THR MET ILE THR PRO SER LEU HIS ALA SER THR LEU GLU ASP PRO ARG VAL PRO SER SER ASN SER
         HindIII SphI    PstI    SalI    XbaI    BamHI SmaI   KpnI    SacI    EcoRI
```

T18   MCS

```
CCC TCC AGG TCG ACT CTA GAG GAT CCC CGG GTA CCG AGC TCG AAT TCA TCG ATA TTA
CGG AGG TCC AGC TGA GAT CTC CTA GGG GCC CAT GGC TCG AGC TTA AGT AGC TAT AAT
ALA GLU ARG SER THR LEU GLU ASP PRO ARG VAL PRO SER SER ASN SER SER TYR STOP
Pst I    Sal I   Xba I   BamHI SmaI  KpnI    Sac2    EcoRI   ClaI
```

T18

MCS:
HindIII
SphI
PstI
[HincII]
AccI
SalI
XbaI
BamHI
SmaI
KpnI
SacI
EcoRI

pUT18
3023 bp
ori ColEI
bla (Amp^r)

MCS:
PstI
[HincII]
AccI
SalI
XbaI
BamHI
SmaI
KpnI
SacI
EcoRI
ClaI

pUT18C
3017 bp
ori ColEI
bla (Amp^r)

FIGURE 13

pCmAHL1

plac UV5

cyaA' MCS cat Ori p15A

1 384

pT25

plac UV5

cyaA' MCS cat Ori p15A

1 224

Pst I BamH I Kpn I

gctgcagggtcgactctagaggatccccgggtacctaagtaactaagaattc

A A G S T L E D P R V P K stop

pT18

plac UV5

αlacZ cyaA' bla Ori C

225 399

Kpnl MCS

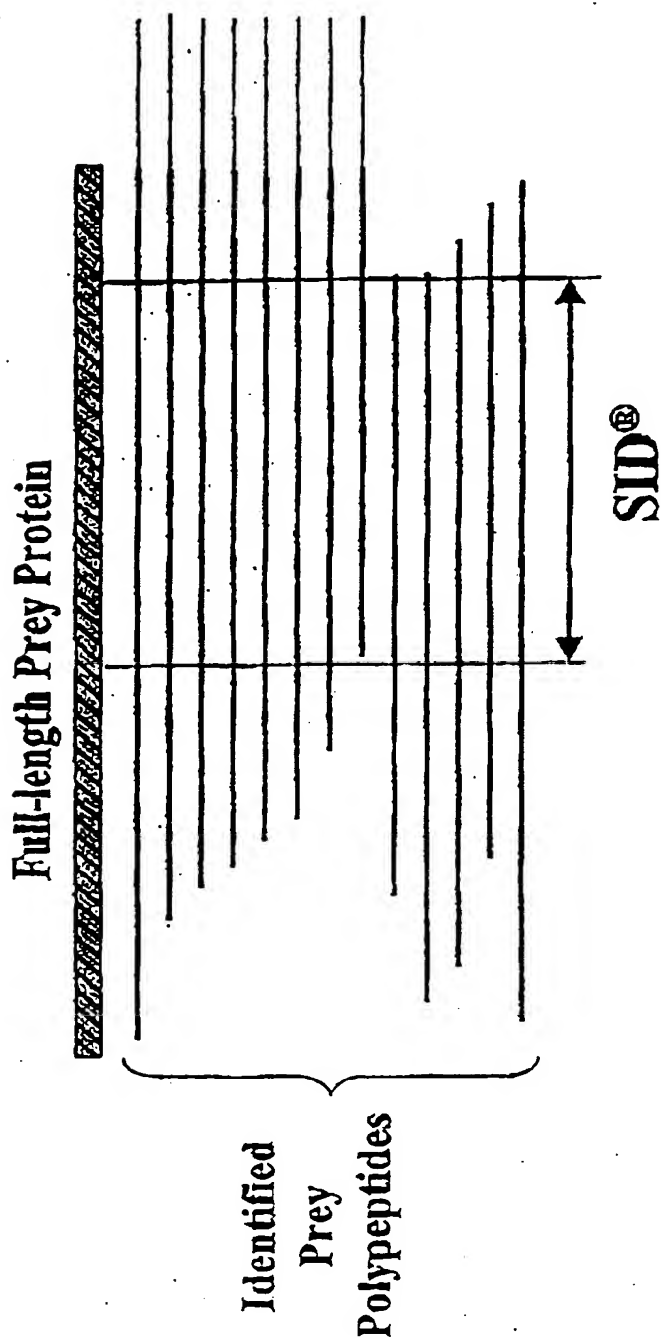tgggtaccgggccccccctcgaggtcgacggtatcgataagcttgatatcgaattcc

W V P G P P S R S T V S I S L I S N S

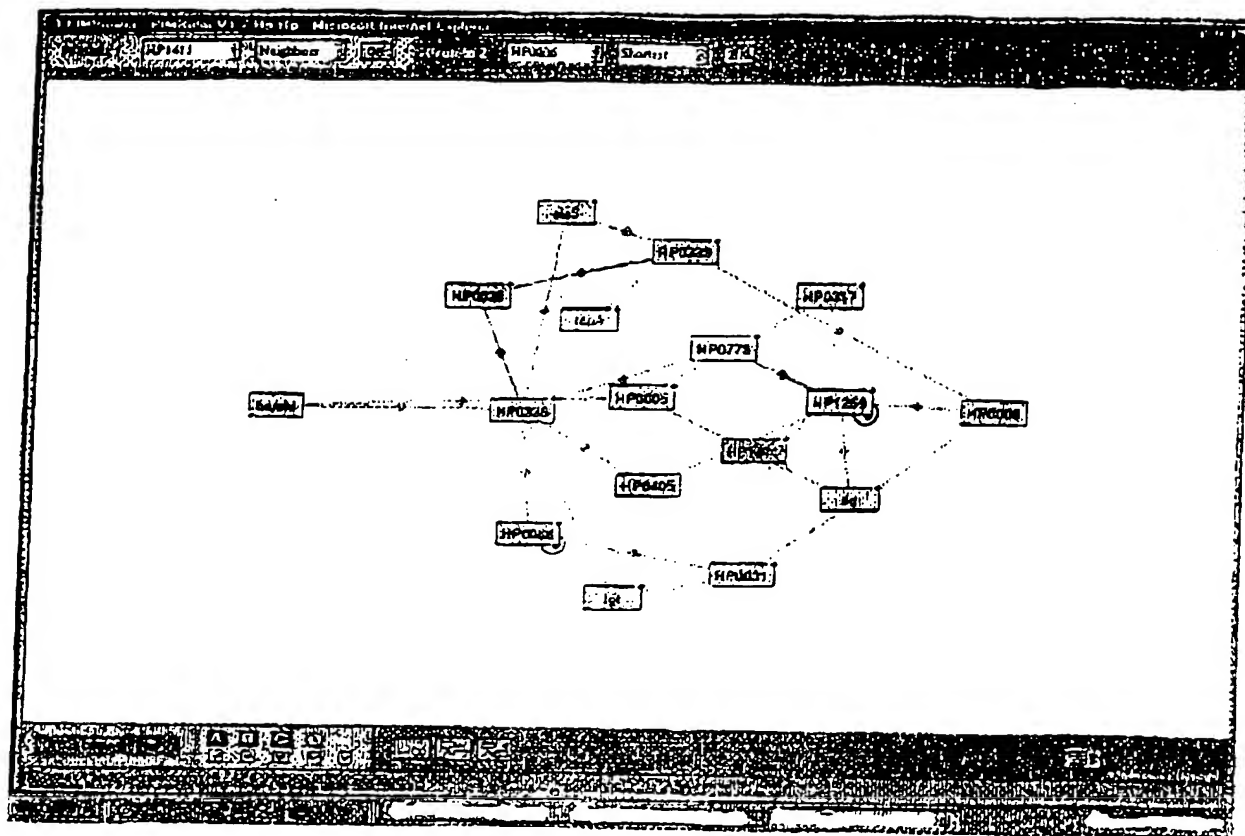FIGURE 14

Figure 15: schematic representation of SID® determination

Figure 16 : Example of Protein Interaction Map